

# THE USE OF CNIDOCYSTS FOR ECOLOGICAL RACES IDENTIFICATION FROM SEA ANEMONES POPULATIONS (ANTHOZOA, ACTINIIDAE)

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## ABSTRACT

The cnidocysts spirocysts, microbasic b-mastigophore, microbasic p-mastigophore and atrichs from tentacles, column and acrorhagi of *Phymactis clematis* Dana, 1849, *Aulactinia marplatensis* (Zamponi, 1977), *A. reynaudi* (M.-Edwards, 1857) and *Oulactis muscosa* Dana, 1849 (Actiniidae) from three intertidal zones of Argentine shore are studied. Treatment of data were made on 5400 cnidocysts by mean an discriminant analysis (BMDP computer programs). Three groups (ecological races) for each species according to the zone were identified, and the results were statistically significant. The microbasic b-mastigophore has the highest variability within them, being the more important type to recognition ecological races.

KEYWORDS. Actiniaria, cnidocysts, variation, intertidal, Argentine shore.

## INTRODUCTION

The variation in size of cnidocysts has been used as a tool in the description of species (CARLGREN, 1940, 1945; HAND, 1961). WEILL (1934) considered that the influence of exogenous and endogenous factors can affect the distribution and size of cnidocysts. ZAMPONI & A.-TELLECHEA (1988) demonstrated that some nematocysts (desmoneme, stenotele and heterotrichous anisorhiza) have a paralyzing function and consequently are larger. FAUTIN (1988) suggested a specific alternative study of the origin of the samples to determine if such variability is caused by external factors. ZAMPONI & ACUÑA (1991) observed for *Actinostola cassicornis* Hertwig, 1882, *Carcinactis dolosa* Riemann-Zürneck, 1975, *Actinauge longicornis* Verrill, 1882, *Bolocera kerguelensis* Studer, 1979, and *Choriactis laevis* Carlgren, 1899, clinal variation in cnidocysts size.

The variation allied with other morphological and ecological characteristics would

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permit the recognition of several actiniarian populations. For this reason we provide a comparative study of the cnidocyst size of four Actiniidae species.

## MATERIALS AND METHODS

The specimens of *Phymactis clematis* Dana, 1849, *Aulactinia marplatensis* (Zamponi, 1977), *A. reynaudi* (M.-Edwards, 1857) and *Oulactis muscosa* Dana, 1849 were taken from July 1990 to July 1992 in the intertidal areas of the beaches Punta Cantera and Punta Piedras in Mar del Plata (38°05'S, 57°32'W) and from Santa Clara del Mar (37°50'S, 57°30'W), Argentina. Punta Cantera and Punta Piedras have a quartzitic substrate where there are exposed and protected areas inhabiting by several benthic organisms such as sponges, cnidarians, molluscs, nemerteans, polychaetes and crustaceans. In Santa Clara del Mar the beach consists of a 5 m high embankment and a gently shelving floor with drainage channels running perpendicular to the coast, and has an intertidal endolithic fauna associated to a compact sedimentary rocks, sometimes cemented by crystalline calcium carbonate, and is of variable colour and hardness. In the three locations the sea rises 50 cm up, the embankment at normal high tide, but under southeast storm conditions can completely cover it.

One hundred cnidocysts of each kind (total= 5400) were measured using the methodology of ZAMPONI & ACUÑA (1994). The cnidocysts were prepared according to HAND's (1954) technique and classified according to ENGLAND (1991). The abbreviations of cnidocysts were (1) tentacle: bmt, microbasic b-mastigophore; et, spirocyst; (2) column: ac, atrichs; bmc, microbasic b-mastigophore; pmc, microbasic p-mastigophore; acrorhagi: aa, atrichs; bma, microbasic b-mastigophore.

A discriminant statistical analysis for each species was done taking into consideration the three areas of the study and the variables were the different cnidocysts. Program 7M of the BMDP package of programs was used. The significance level used was 0.05.

## RESULTS

*Phymactis clematis*. The coefficients of correlation between cnidocysts were: et-bmt -0.00501; et-pmc 0.04590; et-bmc -0.03882; bmt-pmc -0.06565; bmt-bmc -0.00879; pmc-bmc 0.01514.

Were observed statistically significant differences (table I) between individuals of Punta Cantera and Punta Piedras ( $F= 155.04$ ); Punta Cantera and Santa Clara del Mar ( $F= 149.96$ ); but not between Punta Piedras and Santa Clara del Mar ( $F= 4.69$ ), thus, both could be considered as only one group. Individuals from each area represented according to the canonical variables obtained by the discriminant analysis (fig. 1).

According to the value of  $F$  for each one of the cnidocysts, the one that best differentiates the group is microbasic b-mastigophore of tentacle ( $F=156.76$ ) followed in descending order by microbasic p-mastigophore of the column ( $F=127.72$ ), microbasic b-mastigophore of column ( $F=95.08$ ) and spirocyst of tentacle ( $F=20.72$ ).

The following are the discriminant functions for each group which allow a new member to be classed in a group according to the value of its variables, in this case, the size of cnidocysts:  $L(X)pc= 0.3273X_1 + 4.4000X_2 + 2.9009X_3 + 2.9896X_4 - 86.6000$ ,  $L(X)pp= 0.4522X_1 + 5.7047X_2 + 3.6676X_3 + 3.7500X_4 - 139.0103$ ,  $L(X)sc= 0.4171X_2 + 5.4951X_2 + 3.7731X_3 + 3.8604X_4 - 137.7480$ ; where  $X_1= et$ ,  $X_2= bmt$ ,  $X_3= pmc$ ,  $X_4= bmc$ .

*Aulactinia marplatensis*. The coefficients of correlation between cnidocysts were: et-bmt 0.02339; et-pmc 0.02276; et-ac 0.06308; bmt-pmc 0.08881; bmt-ac 0.02106; pmc-ac: 0.06828.

Were observed statistically significant differences between individuals of all zones: Punta Cantera and Punta Piedras ( $F= 183.13$ ); Punta Cantera and Santa Clara del Mar ( $F= 211.67$ ); Punta Piedras and Santa Clara del Mar ( $F= 65.60$ ) (table II). Individuals from each area according to the canonical variables obtained from the discriminant analysis (fig. 2).

The cnidocyst that defines the group better is microbasic b-mastigophore of the

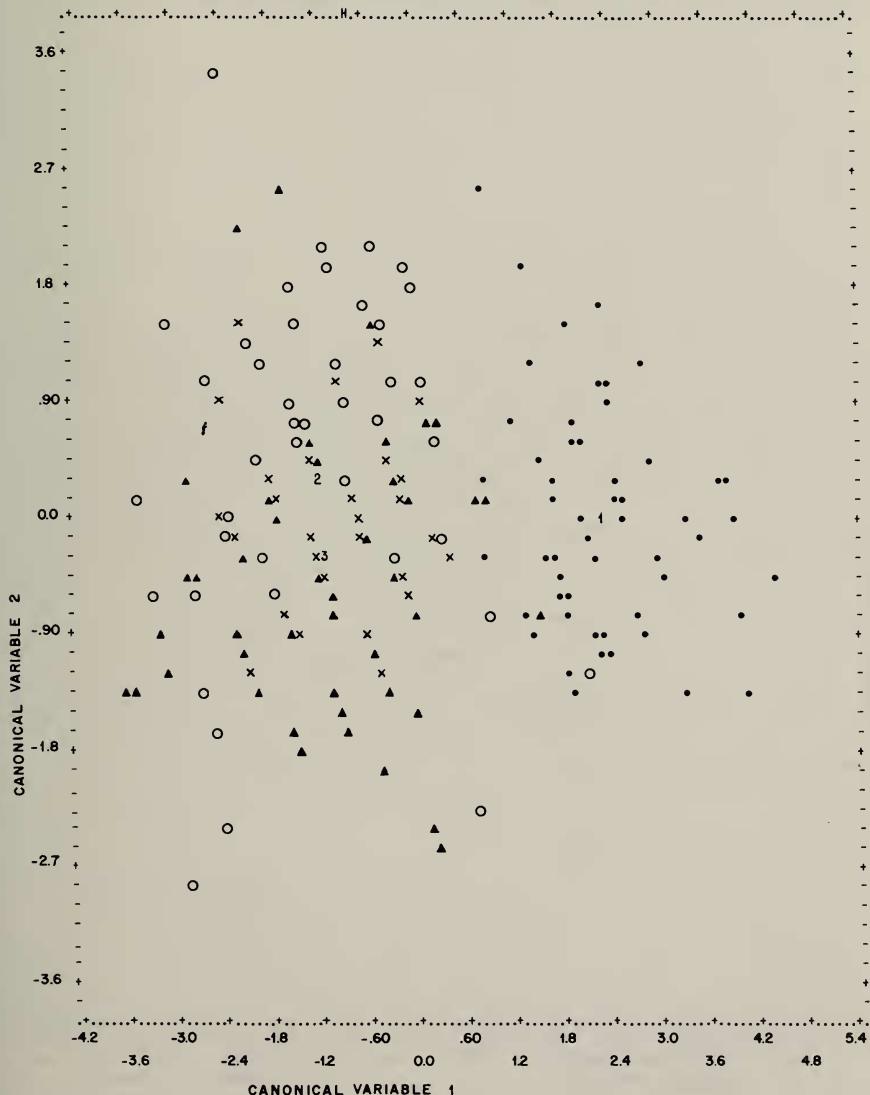


Fig. 1. Individuals of *Phymactis clematis* in the different zones according with cnidocysts size. ( ●, Punta Cantera; ○ Punta Piedras; ▲, Santa Clara del Mar; 1, 2, 3, groups centers; X, superposed individuals).

tentacle ( $F=216.22$ ) followed by microbasic b-mastigophore of the column ( $F=164.82$ ), aitchs of the column ( $F=151.50$ ) and spirocyst of the tentacle ( $F=147.01$ ).

The discriminant functions for each group are the following:  $L(X)pc = 3.7097X_1 + 4.5110X_2 + 3.5791X_3 + 1.1352X_4 - 112.2118$ ,  $L(X)pp = 4.7533X_1 + 5.8880X_2 + 2.5036X_3 + 1.0368X_4 - 170.2202$ ,  $L(X)sc = 4.6145X_1 + 5.6286X_2 + 4.7188X_3 + 1.5080X_4 - 181.9610$ ; where  $X_1 = et$ ,  $X_2 = bmt$ ,  $X_3 = bmc$ ,  $X_4 = ac$ .

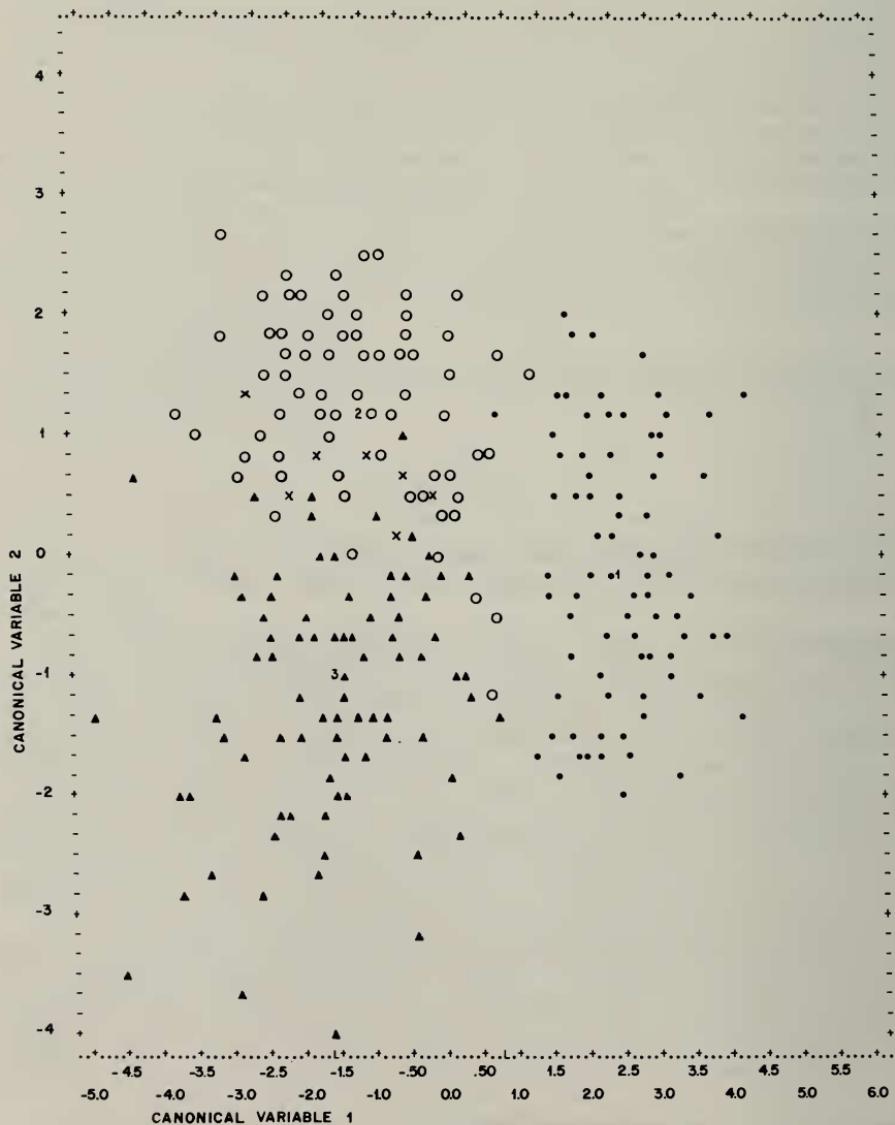


Fig.2. Individuals of *Aulactinia marplatensis* in the different zones according with cnidocysts size. (●, Punta Cantera; ○ Punta Piedras; ▲, Santa Clara del Mar; 1, 2, 3, groups centers; X, superposed individuals).

*Aulactinia reynaudi*. The coefficients of correlation between cnidocyst were: et-bmt 0.08313; et-bmc 0.12476; et-ac -0.04053; bmt-bmc 0.03435; bmt-ac 0.07077; bmc-ac -0.16035. Individuals from the three areas under study have statistical differences: Punta Cantera and Punta Piedras ( $F= 80.06$ ); Punta Cantera and Santa Clara del Mar ( $F= 176.71$ ); Punta Piedras and Santa Clara del Mar ( $F= 50.31$ ) (table III). Individuals according to the canonical variables obtained from the discriminant analysis (fig. 3).

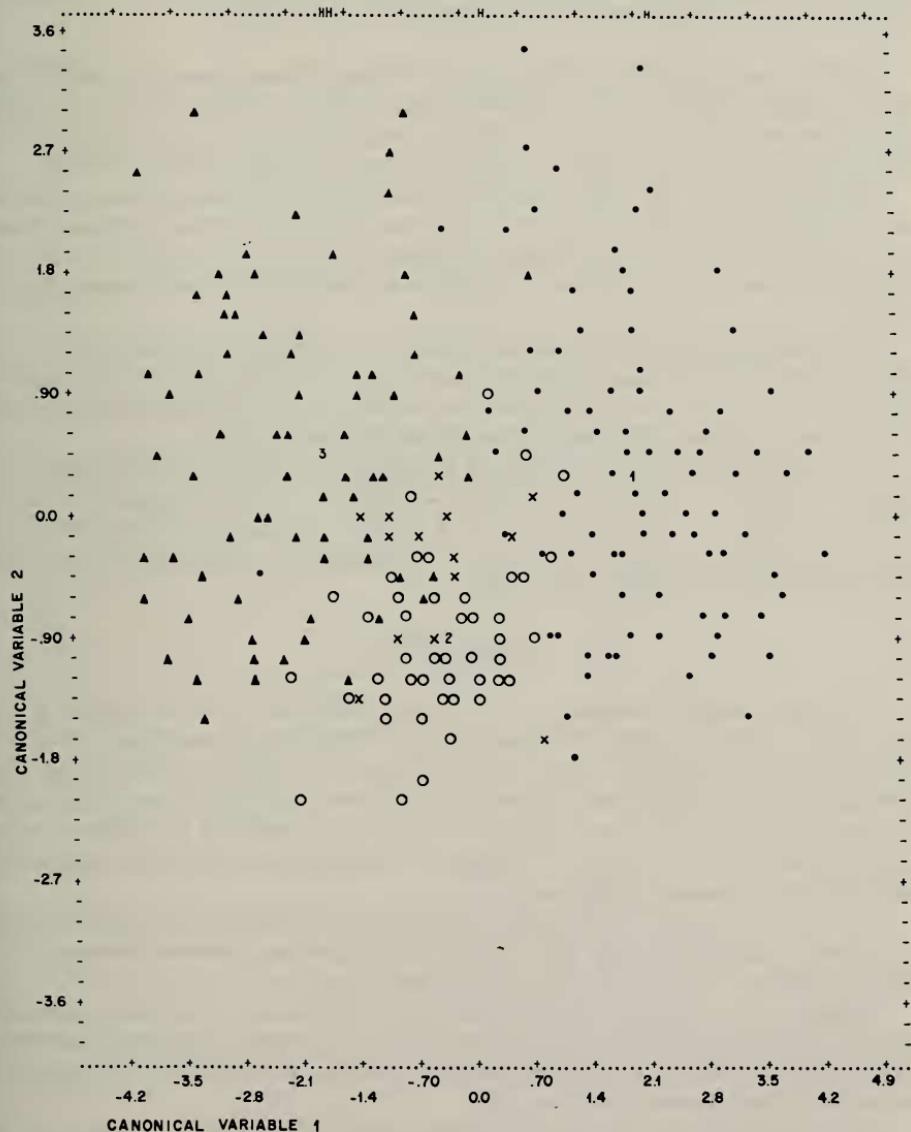


Fig. 3. Individuals of *Aulactinia reynaudi* in the different zones according with cnidocysts size. (●, Punta Cantera; ○, Punta Piedras; ▲, Santa Clara del Mar; 1, 2, 3, groups centers; X, superposed individuals).

The cnidocyst that best defines the group is microbasic b-mastigophore of tentacle ( $F=205.50$ ), then spirocyst of the tentacle ( $F=105.46$ ), microbasic b-mastigophore of the column ( $F=81.03$ ) and atrichs of the column ( $F=62.01$ ).

The discriminant functions for each group are the following:  $L(X)pc = 2.2187X1 + 4.3263X2 + 2.7964X3 + 1.3277X4 - 96.2795$ ,  $L(X)pp = 2.7409X1 + 5.2605X2 + 3.1514X3 + 1.2142X4 - 125.9636$ ,  $L(X)sc = 2.8541X1 + 5.7011X2 + 3.5226X3 + 1.5474X4 - 154.4712$ ; where  $X1 = et$ ,  $X2 = bmt$ ,  $X3 = bmc$ ,  $X4 = ac$ .

*Oulactis muscosa*. The coefficients of correlation between cnidocysts were:  $et-bmt = -0.04975$ ;  $et-bmc = 0.03149$ ;  $et-ac = -0.00740$ ;  $et-aa = -0.02821$ ;  $et-bma = 0.03481$ ;  $bmt-bmc = 0.02113$ ;  $bmt-ac = 0.06273$ ;  $bmt-aa = -0.17257$ ;  $bmt-bma = 0.01382$ ;  $bmc-ac = -0.12021$ ;  $bmc-aa = 0.01957$ ;  $bmc-bma = -0.07735$ ;  $ac-aa = -0.05927$ ;  $ac-bma = -0.04608$ ;  $aa-bma = 0.05229$ .

Individuals from the three zones were statistically different according with the studied variables: Punta Cantera and Punta Piedras: ( $F=80.21$ ), Punta Cantera and Santa Clara del Mar ( $F=83.57$ ), Punta Piedras and Santa Clara del Mar ( $F=121.68$ ) (table IV). Individuals according to the canonical variables obtained from the discriminant analysis (fig. 4).

The cnidocyst that best defines the group is the atrichs of acrorhagi ( $F=273.73$ ), followed by microbasic b-mastigophore of acrorhagi ( $F=141.53$ ), atrichs of the column ( $F=95.55$ ), microbasic b-mastigophore of the tentacle ( $F=39.67$ ), spirocyst of tentacle ( $F=29.43$ ) and microbasic b-mastigophore of the column ( $F=1.51$ ).

The discriminant functions for each group are as follows:  $L(X)pc = 4.609X1 + 6.448X2 + 4.984X3 + 3.506X4 + 3.311X5 + 4.071X6 - 315.885$ ,  $L(X)pp = 5.023X1 + 7.144X2 + 5.195X3 + 3.998X4 + 3.567X5 + 5.166X6 - 388.153$ ,  $L(X)sc = 5.063X1 + 6.587X2 + 5.226X3 + 4.232X4 + 2.814X5 + 4.340X6 - 329.300$ ; where  $X1 = et$ ,  $X2 = bmt$ ,  $X3 = bmc$ ,  $X4 = ac$ ,  $X5 = aa$ ,  $X6 = bma$ .

## DISCUSSION

From the analysis on cnidocysts of *P. clematis* and taking into account the size of these cellular structures, we deduce that this species presents at least three statistically different groups (ecological races), one in each area of study (the three areas were statistically discriminated). However, the separation among individuals from Punta Piedras and Santa Clara del Mar is not significant. Cnidocysts studied of *P. clematis* had no correlation among themselves and microbasic b-mastigophore of tentacle was the one which best discriminated the three areas.

In *A. marplatensis* as in *P. clematis* it was possible to identify individuals according to the size of cnidocysts in the three areas. The differences observed between Punta Cantera and Santa Clara del Mar were particularly important.

Equally, *A. reynaudi* is discriminated in the three areas of the study, especially between individuals from Punta Cantera and Santa Clara del Mar. The cnidocysts of this species had no correlation among themselves. Microbasic b-mastigophore of tentacle is the one which best discriminates the group.

In *O. muscosa*, also, the perfect discrimination of the groups was observed. The cnidocysts do not have any correlation among themselves. Atrichs of acrorhagi was the best to discriminate the groups.

The differences observed between size cnidocysts of different species are not a consequence of a clinal variation. Clinal variations have been observed in actiniarians by ZAMPONI & ACUÑA (1991) but these variations are gradual and occur in very large areas as stated by LAURENT (1972). This was not observed in the studied areas. It is possible that the variation in these cellular structures occurs due to the different conditions, such as olaje and substrate, that prevail in each area. This confirmed the

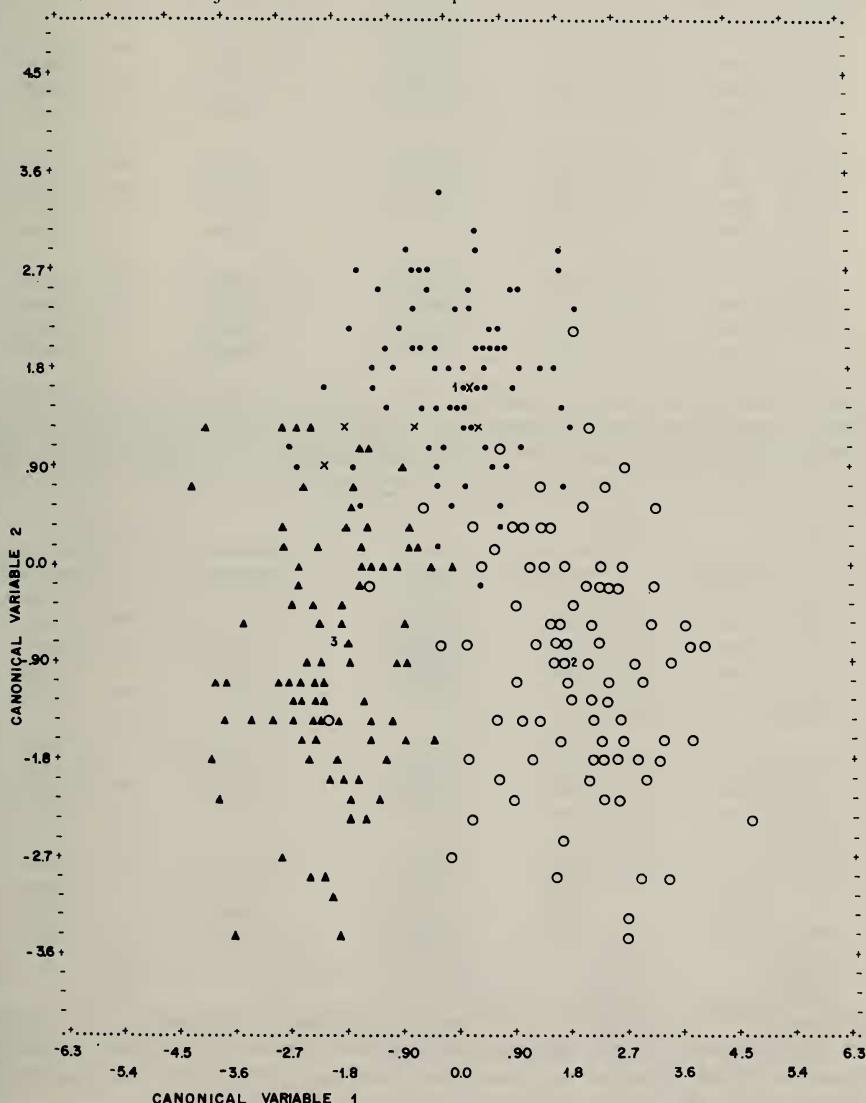


Fig. 4. Individuals of *Oulactis muscosa* in the different zones according with cnidocysts size. (●, Punta Cantera; ○ Punta Piedras; ▲, Santa Clara del Mar; 1, 2, 3, groups centers; X, superposed individuals).

Table I. Statistical data of the variables (cnidocyst) of *P. clematis* in the studied zones. PC, Punta Cantera; PP, Punta Piedras; SC, Santa Clara del Mar. (et, spirocyst of tentacle; bmt, microbasic b-mastigophore of tentacle; pmc, microbasic p-mastigophore of column; bmc=microbasic b-mastigophore of column).

| Zone | Cnidocyst | Number | Mean     | Standard deviation | Coefficient of variation |
|------|-----------|--------|----------|--------------------|--------------------------|
| PC   | et        | 100    | 16.04800 | 11.45331           | 0.71369                  |
|      | bmt       | 100    | 17.56800 | 1.70151            | 0.09685                  |
|      | pmc       | 100    | 16.84800 | 1.53323            | 0.09685                  |
|      | bmc       | 100    | 12.76800 | 1.40151            | 0.10977                  |
| PP   | et        | 100    | 22.15300 | 2.66506            | 0.12030                  |
|      | bmt       | 100    | 22.37900 | 2.28581            | 0.10214                  |
|      | pmc       | 100    | 21.31500 | 3.14750            | 0.14767                  |
|      | bmc       | 100    | 15.99100 | 2.33546            | 0.14605                  |
| SC   | et        | 100    | 20.49900 | 2.46140            | 0.12007                  |
|      | bmt       | 100    | 21.46900 | 2.09288            | 0.09748                  |
|      | pmc       | 100    | 22.01500 | 2.48943            | 0.11308                  |
|      | bmc       | 100    | 16.50300 | 2.34901            | 0.14234                  |

Table II. Statistical data of the variables (cnidocyst) of *A. marplatensis* in the studied zones. PC, Punta Cantera; PP, Punta Piedras; SC, Santa Clara del Mar. (et, spirocyst of tentacle; bmt, microbasic b-mastigophore of tentacle; bmc, microbasic b-mastigophore of column; ac, atrichs of column).

| Zone | Cnidocyst | Number | Mean     | Standard deviation | Coefficient of variation |
|------|-----------|--------|----------|--------------------|--------------------------|
| PC   | et        | 100    | 16.36800 | 1.57319            | 0.09611                  |
|      | bmt       | 100    | 16.88000 | 1.56528            | 0.09273                  |
|      | bmc       | 100    | 14.32000 | 1.14557            | 0.08000                  |
|      | ac        | 100    | 30.04600 | 5.02920            | 0.16738                  |
| PP   | et        | 100    | 20.72700 | 2.22229            | 0.10722                  |
|      | bmt       | 100    | 21.89400 | 1.86156            | 0.08503                  |
|      | bmc       | 100    | 17.86000 | 2.13054            | 0.11929                  |
|      | ac        | 100    | 29.29700 | 2.72639            | 0.09306                  |
| SC   | et        | 100    | 20.44100 | 2.17237            | 0.10628                  |
|      | bmt       | 100    | 21.15300 | 2.06091            | 0.09743                  |
|      | bmc       | 100    | 18.76800 | 2.04993            | 0.10922                  |
|      | ac        | 100    | 39.64000 | 5.75707            | 0.14523                  |

variations observed by PÉREZ (1992) in the epithelial microanatomy of *P. clematis* from the same areas study. Moreover, this author observed statistically significant differences in the variation of some structures (vesicles, tentacles, mesenteries) of taxonomic value for *P. clematis*, *A. marplatensis* and *A. reynaudi*.

The term race does not imply a special taxonomic meaning, but it can be considered as a group of common terms of specific subgroups, such as "variety", "form" and "stock".

Table III. Statistical data of the variables (cnidocyst) of *A. reynaudi* in the studied zones. PC, Punta Cantera; PP, Punta Piedras; SC, Santa Clara del Mar. (et, spirocyst of tentacle; bmt, microbasic b-mastigophore of tentacle; bmc, microbasic b-mastigophore of column; ac, atrichs of column).

| Zone | Cnidocyst | Number | Mean     | Standard deviation | Coefficient of variation |
|------|-----------|--------|----------|--------------------|--------------------------|
| PC   | et        | 100    | 16.43200 | 2.44779            | 0.14897                  |
|      | bmt       | 100    | 17.15200 | 1.60935            | 0.09383                  |
|      | bmc       | 100    | 14.57600 | 3.41799            | 0.23449                  |
|      | ac        | 100    | 29.32800 | 5.96092            | 0.20325                  |
| PP   | et        | 100    | 20.26500 | 1.99597            | 0.09849                  |
|      | bmt       | 100    | 20.57300 | 1.64125            | 0.07978                  |
|      | bmc       | 100    | 17.17000 | 1.23603            | 0.07199                  |
|      | ac        | 100    | 26.23300 | 2.39431            | 0.09127                  |
| SC   | et        | 100    | 21.21600 | 2.87573            | 0.13555                  |
|      | bmt       | 100    | 22.42000 | 2.26845            | 0.10118                  |
|      | bmc       | 100    | 18.66900 | 1.63285            | 0.08746                  |
|      | ac        | 100    | 33.99800 | 5.71457            | 0.16809                  |

Table IV. Statistical data of the variables (cnidocyst) of *O. muscosa* in the studied zones. PC, Punta Cantera; PP, Punta Piedras; SC, Santa Clara del Mar; (et, spirocyst of tentacle; mbt, microbasic b-mastigophore of tentacle; bmc, microbasic b-mastigophore of column; ac, atrichs of column; aa, atrichs of acrorhagi; mba, microbasic b-mastigophore of acrorhagi).

| Zone | Cnidocyst | Number | Mean     | Standard deviation | Coefficient of variation |
|------|-----------|--------|----------|--------------------|--------------------------|
| PC   | et        | 100    | 18.87700 | 1.99311            | 0.10558                  |
|      | mbt       | 100    | 20.92100 | 1.78842            | 0.08548                  |
|      | bmc       | 100    | 19.58800 | 1.50207            | 0.07668                  |
|      | ac        | 100    | 19.34500 | 1.42058            | 0.07343                  |
|      | aa        | 100    | 52.97200 | 4.39290            | 0.08293                  |
|      | mba       | 100    | 16.39900 | 1.35023            | 0.08234                  |
| PP   | et        | 100    | 20.63400 | 1.88911            | 0.09155                  |
|      | mbt       | 100    | 23.38800 | 2.22517            | 0.09514                  |
|      | bmc       | 100    | 19.99300 | 2.41202            | 0.12064                  |
|      | ac        | 100    | 22.26200 | 1.91051            | 0.08582                  |
|      | aa        | 100    | 56.93700 | 4.74615            | 0.08336                  |
|      | mba       | 100    | 20.91700 | 2.80321            | 0.13466                  |
| SC   | et        | 100    | 20.98000 | 2.32800            | 0.11096                  |
|      | mbt       | 100    | 22.37600 | 1.86554            | 0.08337                  |
|      | bmc       | 100    | 20.05800 | 2.19348            | 0.10936                  |
|      | ac        | 100    | 24.43200 | 3.84584            | 0.15741                  |
|      | aa        | 100    | 42.80400 | 3.94710            | 0.09221                  |
|      | mba       | 100    | 17.06900 | 1.52830            | 0.08954                  |

(WILLIAMS, 1973). It is possible that the ecological races are closely related with physiological races as pointed out by WILLIAMS (1973) for the sea anemone *Haliplanella luciae* Hand, 1955. UCHIDA (1936) described four races in *H. luciae* in

Japan, based on color differences and suggesting that their distribution is influenced by the effect of temperature and ocean currents.

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